

CLAIMS

1. A process for the production of a polyunsaturated fatty acid (PUFA), the
5 process comprising culturing a micro-organism in a culture medium inside a
fermentation vessel, whereby at a stage which precedes the end of fermentation;
 - a) the carbon source is consumed by the micro-organisms at a rate greater
than it is added to the medium;
 - b) the carbon source is added at a rate of $\leq 0.30\text{M}$ carbon/kg medium per
10 hour;
 - c) the carbon source is rate limiting on the growth of the micro-organisms,
or is restricted such that the micro-organisms metabolise fat(s) and/or
lipid(s);
 - d) the rate of addition of the carbon source is reduced or is below the rate of
15 consumption of the carbon source by the micro-organisms; or
 - e) the carbon source has been all used, or has a concentration in the medium
of about zero, at or before the end of fermentation;
 - f) the carbon source addition is stopped but fermentation is allowed to
continue; and/or
 - 20 g) the micro-organisms are subjected to conditions whereby they metabolise,
or consume, one or more fat(s) or lipids(s) in preference to arachidonic
acid (ARA).
2. A process according to claim 1, wherein:
 - h) the concentration of the carbon source in that stage is on average \leq
25 10g/kg and/or $\leq 0.17\text{ M}$ carbon/kg medium;
 - i) the carbon source is glucose; and/or
 - j) the PUFA is present in a microbial oil.
- 30 3. A process according to claim 1 or 2, wherein:
 - k) the second stage starts at from 15 to 2 hours before the end of the
fermentation or less than 10 days after the beginning of the fermentation.

4. A process according to any one preceding claim, wherein:
- 1) the (entire) fermentation is carried out at a temperature $\geq 22^{\circ}\text{C}$ or and/or $\leq 30^{\circ}\text{C}$;
 - 5 m) it is conducted in the absence of an additive oil; and/or
 - n) the fermentation lasts no longer than 9 days.

5. A process according to any preceding claim wherein:
- 10 n) the PUFA comprises arachidonic acid (ARA); and/or
 - o) the vessel has a capacity of at least 10 litres.

6. A process according to any preceding wherein the micro-organism is *Mortierella*, optionally *Mortierella alpina*, and/or is non-genetically modified.

- 15 7. A microbial oil which comprises at least 50% arachidonic acid (ARA) and:
- a) has at least a 90% triglyceride content;
 - b) has a peroxide value (POV) of no more than 2.5;
 - c) has an anisidine value (AnV) of no more than 1.0;
 - d) is hexane extracted; and/or
 - 20 e) has a phospholipid content below 5%.

8. An oil according to claim 7 which comprises:
- f) less than 5% of C₂₀ and/or C₂₄ polyunsaturated fatty acids (PUFAs); and/or
 - 25 g) less than 5% of C₂₂₊ PUFAs.

9. An oil according to claim 7 or 8 wherein:
- h) the free fatty acid content is $\leq 0.4\%$;
 - i) the triglyceride content is at least 95% or 98%; and/or
 - 30 j) the oil is preparable by a process according to any one of claims 1 to 6.

10. A composition comprising a microbial oil obtainable by a process according to any one of claims 1 to 6 and/or a microbial oil according to any one of claims 7 to 9.

11. A composition according to claim 10 which is a foodstuff (such as an infant

5 formula), food, feed, or feed supplement, pharmaceutical, veterinary or cosmetic composition.